OPEM
www.opem.org

Oriental Pharmacy and Experimental Medicine 2010 **10(2)**, 59-65
DOI 10.3742/OPEM.2010.10.2.059

Antihyperglycemic and lipid lowering effect of *Tectona grandis* in alloxan induced diabetic rats

Ghaisas MM[1,*], Navghare VV[2], Takawale AR[2], Zope VS[2], Tanwar MB[2] and Phanse MA[2]

[1]Principal, India College of Pharmacy, Pune, Indira; [2]Department of Pharmacology Padm. Dr. D. Y. Patil Institute of Pharmaceutical Sciences and Research, Pimpri, Pune-18, Indira

Received for publication January 06, 2008; accepted May 06, 2010

SUMMARY

In India, *Tectona grandis* is traditionally used in the treatment of diabetes mellitus and lipid disorder. In the present study, the antihyperglycemic and lipid lowering effect of ethanolic extract of bark of *Tectona grandis* (TG) was evaluated using alloxan induced diabetes in rats. Alloxan was given at the dose of 140 mg/kg, i.p. After induction of diabetes, TG was administered for 42 days p. o. and simultaneously different biochemical parameters like plasma glucose, liver glycogen content, serum triglyceride, cholesterol, LDL-cholesterol and HDL-cholesterol were estimated. Diabetic control showed significant increase ($P < 0.01$) in plasma glucose, serum triglyceride, cholesterol, LDL-cholesterol and significant decrease ($P < 0.01$) in serum HDL-cholesterol and liver glycogen content. Treatment with TG showed significant reduction ($P < 0.01$) in plasma glucose when compared with diabetic control. The elevated levels of serum triglyceride and cholesterol levels were significantly reduced ($P < 0.01$) by TG. TG treatment for 42 days showed significant decrease in serum LDL-cholesterol ($P < 0.01$) and significant increase in serum HDL-cholesterol level ($P < 0.01$). Moreover, diabetic control there was significant decrease in liver glycogen content which was significantly increased ($P < 0.05$) by treatment with TG. Hence, from the result obtained in the present study it can be concluded that *Tectona grandis* has the potential to treat diabetes condition and associated lipid disorder.

Key words: *Tectona grandis;* Alloxan; Antihyperglycemic; Lipid lowering; Glucose; Glycogen

INTRODUCTION

Diabetes mellitus ranks highly among the top ten disorders which cause mortality throughout the world. Diabetes mellitus is associated with hyperlipidemia and co-morbidities such as obesity and hypertension. Hyperlipidemia is a metabolic complication of both clinical and experimental diabetes. Diabetes mellitus being chronic disorder, treatment without side effects for long term control is important. Diabetes mellitus is not a single disease entity, but rather a group of metabolic disorders sharing a common underlying feature of hyperglycemia. Hyperglycemia in diabetes, results from defects in insulin secretion, insulin action or most commonly both (Cotran and Robbins, 2004).

Present antidiabetic agents possess side effect like risk of hypoglycemia, anemia, cholestatic jaundice (Goodman and Gilman, 2001). Many plant constituents have varying degree of hypoglycemic and anti-hyperglycemic activity and uptill now no case of adverse effect is counted with plant constituents. Among these are alkaloids, glycosides, galactomannan,

*Correspondence: Ghaisas MM. Principal, Indira College of Pharmacy, Pune, India. Tel: +919422080072; E-mail: ghaisasmm@yahoo.com

polysaccharides, peptidoglycans, hypoglycans, guanidine, steroids, carbohydrates, glycopeptides, terpenoids, amino acids and inorganic ions (Goyal *et al*., 2008).

In India, *Tectona grandis* Linn. is traditionally used in the treatment of diabetes and lipid disorders (The Ayurvedic Pharmacopeia of India, 1996). The ethanolic extract of trunk bark and wood chips of *Tectona grandis* Linn. has antiulcer activity in rats. The ethanolic extract of *Tectona grandis* (TG) consists of triterpenoides, saponin, glycoside, steroids, and tannins (Pandey *et al*., 1982). The hydroalcoholic extract of leaves of *TG* Linn. reported for wound healing potential (Majumdar *et al*., 2007). Chloroform-methanol extract of sawdust of TG Linn. has antifungal activity (Sumthong *et al*., 2006). The petrol extract of root of TG Linn. has cytotoxic activity (Khan and Miungwana, 1999). On the basis of reported activities and chemical constituents the ethanolic extract was chosen. Therefore, taking into consideration the reported pharmacological activities of TG Linn. the present study is an effort to validate scientifically antihyperlipidemic potential of TG in alloxan induced diabetic rats.

MATERIALS AND METHODS

Plant material and preparation of extract

The bark of TG Linn. (Verbenaceae) was collected in the month of June 2007. The plant was authenticated by Dr. A. M. Mujumdar (Head, Plant Sciences Division) Agharkar Research Institute; Pune as TG Linn. (Verbenaceae) with a voucher specimen no. Auth08-012.

The bark was washed with distilled water, shed dried and latter powdered. This powder was then defatted with petroleum ether which was then macerated with ethanol for 72 h with occasional shaking. It was then filtered and the solvent was evaporated under vacuum. The yield of ethanolic extract of TG Linn. was 2.7% w/w.

TG when subjected for phytochemical study showed the presence of beta-sitosterol, terpenoids, phenolic compounds, saponins, glycosides and tannins (Khandelwal, 2005).

Animals

Wistar rats weighing 160 - 200 g and swiss albino mice weighing 20 - 30 g were obtained from National Toxicology Center (NTC), Pune and were housed under standard laboratory conditions. (22 ± 3°C temperature, relative humidity 30%, 12h light and dark cycle). Animals had free access to standard pellet diet and water *ad libitum*. The Institutional Animal Ethics Committee approved the experimental protocol (IAEC registration no. 198/99/CPCSEA).

Experimental design

Acute toxicity study (OECD 425, 2001)

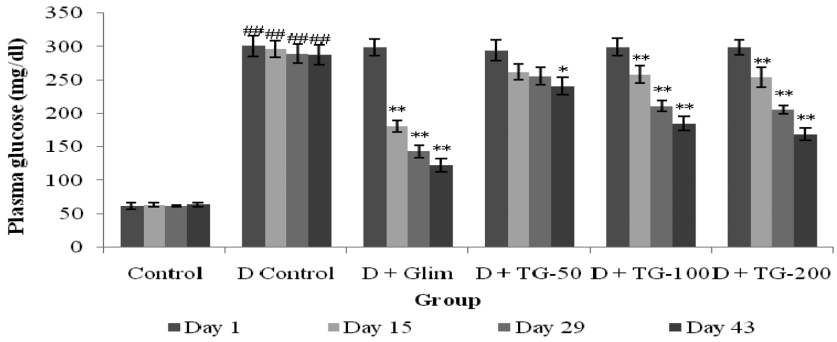

Fig. 1. Effect of TG Linn. on plasma glucose level in alloxan induced diabetic rats.
D Control, diabetic control; TG, ethanolic extract of TG Linn. 50, 100 and 200 mg/kg, p.o. Values are expressed as Mean ± SEM. (n = 5), ANOVA followed by Dunnett test. $^{\#\#}P < 0.01$ when compared with Control; $^{*}P < 0.05$, $^{**}P < 0.01$ when compared with Diabetic control.

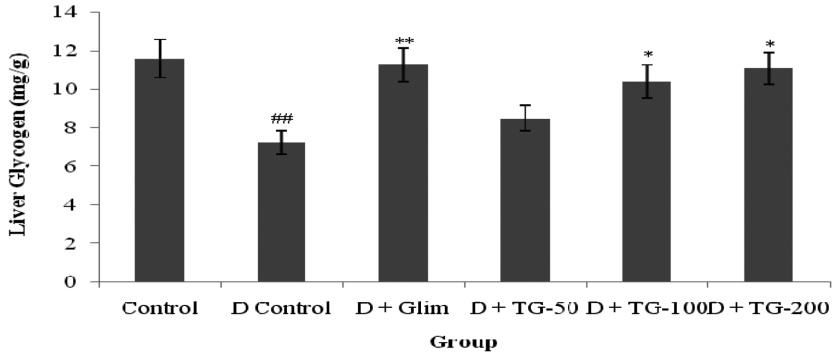

Fig. 2. Effect of TG Linn. on liver glycogen content in alloxan-induced diabetic rats.
D Control, diabetic control; TG, ethanolic extract of TG Linn. 50, 100 and 200 mg/kg, p.o. Values are expressed as Mean ± SEM. (n = 5), ANOVA followed by Dunnett test. $^{\#\#}P < 0.01$ when compared with Control; $^{*}P < 0.05$, $^{**}P < 0.01$ when compared with Diabetic control.

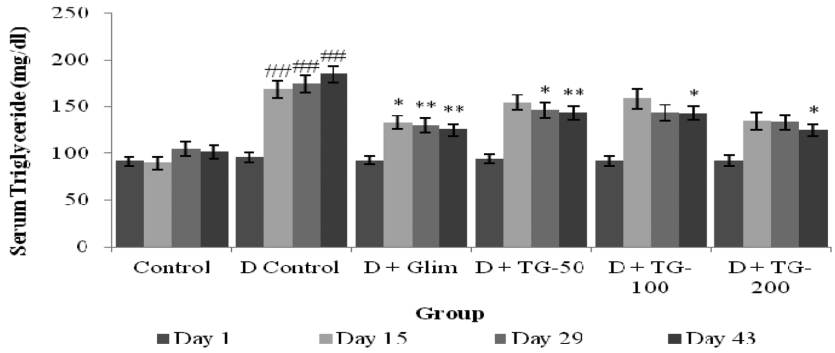

Fig. 3. Effect of TG Linn. on serum triglyceride level in alloxan induced diabetic rats.
D Control, diabetic control; TG, ethanolic extract of *Tectona grandis* Linn. 50, 100 and 200 mg/kg, p.o. Values are expressed as Mean ± SEM. (n = 5), ANOVA followed by Dunnett test. $^{\#\#}P < 0.01$ when compared with Control; $^{*}P < 0.05$, $^{**}P < 0.01$ when compared with Diabetic control.

Acute toxicity of ethanol extract of TG was done using Swiss albino mice (25 - 30 g) according to the procedure of Organization for Economic Co-operation and Development (OECD) guideline no. 425 (OECD, 2001). The animals were fasted overnight prior to the experiment and maintained under standard conditions. TG was found safe up to dose of 2,000 mg/kg, p.o.

Induction of diabetes

Diabetes was induced by single intraperitonial injection of alloxan monohydrate in citrate buffer (pH 4.5) at a dose of 140 mg/kg, body weight of the rat. (Ghosh and Suryawanshi, 2001; Reshmi *et al*., 2001; Murali *et al*., 2002; Prince *et al*., 2004). The diabetic state was confirmed 48 h after alloxan injection by hyperglycemia. Surviving animals with fasting blood glucose level higher than 250 mg/dl were included in the study.

Treatment schedule

Total of 30 Wistar rats were used (25 Diabetic Surviving and 05 nondiabetics). The rats were divided in to 06 groups (n = 5) as follows-The solution of TG was prepared with 1% gum acacia, an emulsifying agent. Glimepride was served as a reference standard. Animals were divided into following groups.

Group-I (Control) animals were non-diabetics and received only 1% gum acacia (1 ml/kg/day,

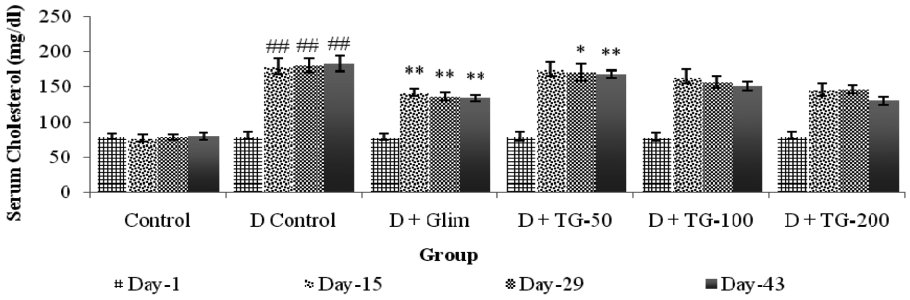

Fig. 4. Effect of TG Linn. on serum Cholesterol level in alloxan induced diabetic rats.
D Control, diabetic control; TG, ethanolic extract of TG Linn. 50, 100 and 200 mg/kg, p.o. Values are expressed as Mean ± SEM. (n = 5), ANOVA followed by Dunnett test. $^{\#\#}P < 0.01$ when compared with Control; $^{*}P < 0.05$, $^{**}P < 0.01$ when compared with Diabetic control.

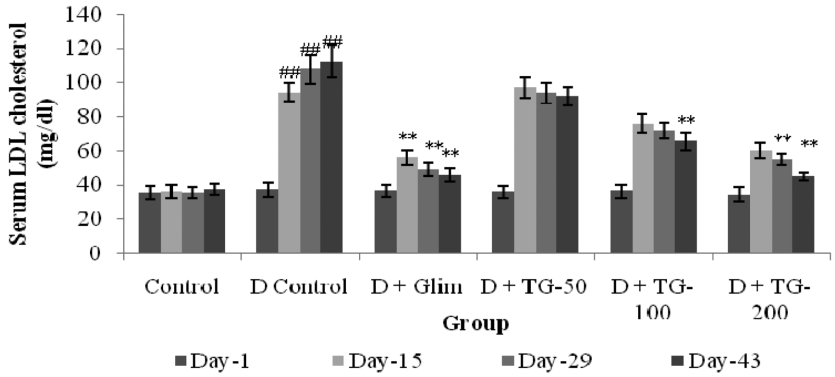

Fig. 5. Effect of TG Linn. on serum LDL-Cholesterol level in alloxan induced diabetic rats.
D Control, diabetic control; TG, ethanolic extract of TG Linn. 50, 100 and 200 mg/kg, p.o. Values are expressed as Mean ± SEM. (n = 5), ANOVA followed by Dunnett test. $^{\#\#}P < 0.01$ when compared with Control; $^{*}P < 0.05$, $^{**}P < 0.01$ when compared with Diabetic control.

p.o.) for six weeks. Group-II (D-Control) animals were diabetic and received 1 % gum acacia (1 ml/ kg/day, p.o.) for six weeks. Group-III (D + Glim) animals were diabetic and received glimepride (0.09 mg/kg/day, p.o.) for six weeks. Groups IV, V, VI animals were diabetic and received three different doses of TG 50, 100, 200mg/kg, p.o. respectively for six weeks.

Biochemical parameters from blood

Blood sample of each of each animal were collected under light ether anesthesia from the retro-orbital plexus on day 1 and at the end of every week of treatment for estimation of plasma glucose (GOD/ POD Method), total cholesterol (COD/POD Method), triglyceride (GPO/POD Method), HDL-cholesterol, LDL-cholesterol.

Study of morphometric parameters

Body weight was recorded through out the study period.

Determination of glycogen content in liver

On last day, rats were sacrificed by cervical dislocation. Liver samples were dissected out and washed immediately with ice cold saline to remove as much blood as possible. Liver homogenates (5% w/v) were prepared in cold 50 mM Tris buffer (pH 7.4) using Remi homogenizer. The unbroken cells and cell debris was removed by

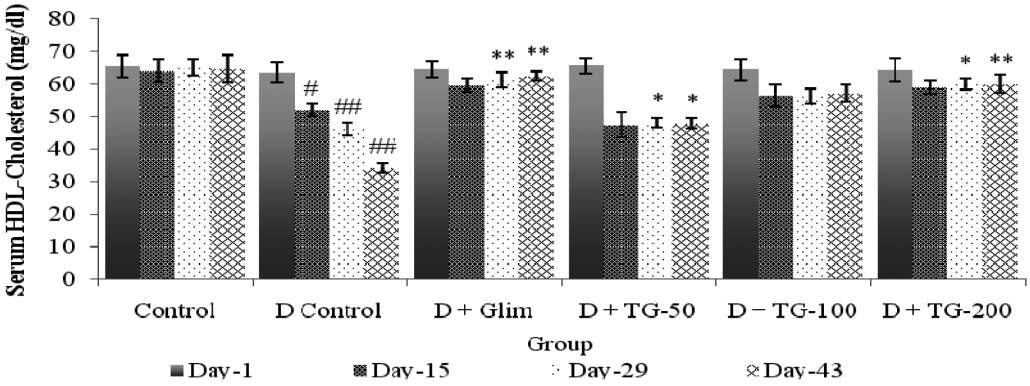

Fig. 6. Effect of TG Linn. on serum HDL-Cholesterol level in alloxan induced diabetic rats.
D Control, diabetic control; TG, ethanolic extract of TG Linn. 50, 100 and 200 mg/kg, p.o. Values are expressed as Mean ± SEM. (n = 5), ANOVA followed by Dunnett test. $^{\#\#}P < 0.01$ when compared with Control; $^{*}P < 0.05$, $^{**}P < 0.01$ when compared with Diabetic control.

centrifugation at 5000 rpm for 10 min using a Remi refrigerated centrifuge. The liver glycogen was measured using method previously described. (Carroll *et al*., 1955).

Statistical analysis

The results were expressed as mean ± SEM and statistically analyzed by ANOVA followed by Dunnett test, with level of significance set at $P < 0.05$.

RESULTS AND DISCUSSION

Diabetes mellitus ranks highly among the top ten disorders which cause mortality throughout the world. Diabetes mellitus is associated with hyperlipidemia and co-morbidities such as obesity and hypertension. In the present study, in diabetic control group there was significant increase in the levels of plasma glucose. The diabetic animals treated with TG for 42 days showed significant reduction in the elevated levels of plasma glucose. The antihyperglycemic effect of TG may be due to the chemical constituents of TG like glycosides, terpenoids, tannins, and saponins. Substances like glycosides, terpenoids, tannins and saponins are frequently implicated as having antidiabetic effects (Matsudha *et al*., 2002; Guy *et al*., 2007; Ghaisas *et al*., 2009) and are the major constituents of TG (Khare, 2007).

One possible effect of antidiabetic drugs can be improvement of glycogenesis (Maiti *et al*., 2004). Diabetic control group showed significant decrease in liver glycogen content while treatment with TG for 42 days resulted in significant and dose dependant elevation of the liver glycogen content. This focuses one possible way of antidiabetic action of TG, which is improvement of glycogenesis process in liver.

The plethora of glucose outside the cells in diabetes contrasts with the intracellular deficit. Glucose catabolism is normally a major source of energy for cellular processes. In diabetes, energy requirements can be met only by drawing the protein and fat reserves. In diabetes there is increase in the catabolism of protein and fat which causes the increase in plasma lipid levels (Cotran and Robbins, 2004).

Chronic diabetes is always associated with derangement of lipid metabolism. In diabetes, enhanced activity of the hormone sensitive lipases increases the lipolysis and releases more free fatty acids in the circulation. Increased fatty acid concentration also increases the β-oxidation of fatty acids, producing more acetyl-CoA, and cholesterol during diabetes (Ozturk *et al*., 1998). In the present study, diabetic control showed significant increase

in level of triglycerides, serum cholesterol, LDL-cholesterol and decrease in HDL-cholesterol level. Treatment with TG showed significant decrease in triglyceride level. Saponins also act as antihyperlipidemic by binding with cholesterol in intestinal lumen, so that cholesterol is less readily absorbed and besides increasing lipoprotein lipase activity which helps in removal of LDL and chylomicrons from circulation (Morehouse *et al*., 1999). Diabetic animals treated with TG showed significant and dose dependant reduction in levels of serum cholesterol and LDL-cholesterol, and improvement in HDL-cholesterol levels may act by inhibiting cholesterol synthesis and increase excretion of cholesterol which may probably due to presence of steroids and saponins. Possible mechanism for this lipid lowering activity of TG may be the increase in uptake and utilization of glucose leading to glycemic control and subsequent inhibition of lipolysis.

Hence, the results obtained in the present study indicate that TG has the potential to treat diabetes mellitus and associated complications owing to its antihyperglycemic and lipid lowering effect. Further studies are necessary to substantiate above observation and to work out exact mechanism of action involved in antihyperlipidemic activity of this plant.

REFERENCES

Carroll NV, Longley RW, Roe JH. (1955) The determination of Glycogen in liver and muscle by use of Anthrone reagent. *J. Biol. Chem.* 583-593.

Ghaisas M, Navghare V, Takawale A, Zope V, Tanwar M, Deshpande A. (2009) Effect of *Tectona grandis* Linn. on dexamethasone-induced insulin resistance in mice. *J. Ethnopharmacol.* **122**, 304–307.

Ghosh S, Suryawanshi SA. (2001) Effect of *Vinca rosea* extract in treatment of Alloxan diabetes in male albino rats. *Indian J. Expt. Biol.* **39**, 748-759.

Goyal RK, Mehta AA, Mahajan SG. (2008) Classification of Herbal Antidiabetic Based on Mechanism of Action and Chemical Constituetns. In: Govil JN, Singh VR, Mishara SK. Recent progress in medicinal plants vol 20, Phytopharmacology and Therapeutic values II. USA, Stadium Press LLC, pp. 65-110.

Guy K, Jaekyung K, Klaus H, Yanyan C, Xiaozhuo C. (2007) Antidiabetes and antiobesity activity of *Laserstroemia speciosa*. *eCAM* **10**, 1-7.

Khan RM, Miungwana SM. (1999) 5-Hydroxylapachol: a cytotoxic agent from *Tectona grandis. Phytochemistry* **50**, 439-442.

Khandelwal KR. (2005). Practical Pharmacognosy, XIV ed. pp. 149-153 Nirali Prakashan, Pune.

Khare CP. (2007) *Indian Medicinal Plants*. An Illustrated Dictionary, pp. 649-650, Berlin/Heidelberg. Springer-Verlag.

Maiti R, Jana D, Das UK, Ghosh D. (2004) Antidiabetic effect of aqueous extrat of *Tamarindus indica* in STZ-induced diabetic rats. *J. Ethnopharmacol.* **92**, 85-91.

Majumdar M, Nayeem N, Kamat JV, Asad Md. (2007) Evaluation of *Tectona grandis* Leaves for wound healing activity. *Pakistan J. Pharma. Sci.* **20**, 120-124.

Matsudha H, Morikawa T, Yoshikawa M. (2002) Antidiabetogenic constituents from several natural medicine. *Pure Appl. Chem.* **74**, 1301-1308.

Morehouse L, Ban Gerter FW, Ninno M, Inskeep B, Carthy P, Pethini J. (1999) Comparison of synthetic saponin cholesterol absorption inhibitors in rabbits: evidence for a non-stoichiometric, intestinal mechanism of action. *J. Lipid Res.* **40**, 464-437.

Murali B, Goyal RK. (2002) Effect of chronic treatment with Losartan on Streptozotocin induced diabetic rats. *Indian J. Expt. Biol.* **40**, 31-34.

OECD (2001) *Guidelines for Testing of Chemicals* 425, Acute Oral Toxicity-Up-and-Down Procedure, p. 1-26.

Ozturk Y, Altan VM, Ari N. (1998) Diabetic Complications in Experimental Models. *Tr. J. Med. Sci.* **22**, 331-341.

Pandey BL, Goel RK, Pathak NKR, Biswas M, Das PK. (1982) Effect of *Tectona grandis* Linn. (common Teak tree) on experimental ulcers and gastric secretion. *Indian J. Med. Res*. **76**, 89-94.

Prince PSM, Kamalakkannan N, Menon VP. (2004) Antidiabetic and antihyperlipidaemic effect of alcoholic *Syzigium cumini* seeds in alloxan induced diabetic albino rats. *J. Ethnopharmacol.* **91**, 209–213.

Reshmi CR, Fatima A, Sinilal B and Latha MS. (2001) Antidiabetic effect of herbal drug in Alloxan diabetic rats. *Indian Drugs* **38**, 319-322.

Robbins and Cotran. (2004) The endocrine pancreas. Pathologic basis of disease, pp. 1189-1207, Elsevier India.

Schimmer BP, Parker KL. (2001) Adrenocorticotropic Hormone; Adrenocortical Steroids and their Synthetic Analogs; Inhibitors of Synthesis and Actions of Adrenocortical Hormones. In: Hardman JG, Limbard LE, Gilman AG, Goodman-Gilman A, eds., Goodman & Gilman's The Pharmacological Basis of Therapeutics. p. 1658-1659, McGraw-Hill.

Sumthong P, Damveld RA, Choi YH, Arentshorst M, Ram AFJ, Van den Hondel CAMJJ, Verpoorte R. (2006) Activity of Quinones from Teak (*Tectona grandis*) on Fungal Cell Wall Stress. *Planta Med.* **72**, 943-944.

The Ayurvedic Pharmacopoeia of India. (1996) Part I, Vol. IV, pp. 122-123, The Controllers of Publications Civil lines, Delhi.